tetanus, but the signs were only mild to moderate. In these mice, also, no significant pathological lesions were discovered.

The intraperitoneal injection of either 25 or 2500 units of tetanal antitoxin caused no recognizable clinical or pathological manifestations.

Summary. The administration of 25 units of tetanal antitoxin to white Swiss mice which had previously received 1 M.L.D. of crystalline tetanal toxin, produced no discoverable lesions in the central nervous system, peripheral nerves, or skeletal muscles, irrespective of whether the animals developed clinical tetanus, or of how long they survived. Likewise, the intraperitoneal injection of either 25 or 2500 units of tetanal antitoxin caused no changes which could be detected by ordinary histopathological technics.

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Effect of *Lithospermum ruderale* on the Gonadotropic Potency of the Pituitary Gland.*

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(Introduced by Raymond N. Bieter.)

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The crude plant, *Lithospermum ruderale,* has been shown to cause prolonged or persistent diestrous in mice with previously regular estrous cycles.[1,2] Since the administration of either follicle-stimulating hormone or estrogenic hormone induces estrus in animals under treatment with Lithosperm and since Lithosperm causes a decrease in the weights of pituitary glands the suggestion has been made that the drug acts on the pituitary gland to decrease the formation or secretion of gonadotropic hormone.[1] The present paper deals with the biological assay for gonadotropic potency of pituitary glands from treated and control animals.

Experimental. Female mice, 21 days old, were divided into 3 groups, distributing littermates between the groups as evenly as possible. Group I called *Lithosperm donors,* consisted of 9 mice, each of which was given an injection of two pituitary glands from donor mice which had been on a 20% ground Lithosperm diet for one week or more. In Group II, the control donors, each of 9 mice received an injection of 2 pituitary glands from donor mice on control diet, Purina fox chow. Group III served as uninjected controls, receiving no pituitary injections. Donor mice for Groups I and II were adult females and were sacrificed during diestrus only. To make the pituitary injections a method similar to that of Smith and Engle[3] was used. Donor mice were killed with ether, the skin on back of head calvarium and brain removed and pituitary gland lifted out. Recipient mice were lightly anesthetized with ether and the pituitary glands placed through a small incision into the thigh muscles. Mice were autopsied at 26 days of age, *i.e.* 5 days after injection. The ovaries, uterus plus vagina and adrenal glands were dissected out and weighed in the moist state.

The data obtained are presented in Table I. Evidence of gonadotropic stimulation was

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1 Cranston, E. M., *J. Pharm. and Exp. Therap.,* 1945, **83**, 130.

2 Drasher, M. L., and Zahl, P. A., PROC. SOC. EXP. BIOL. AND MED., 1946, **63**, 66.

3 Smith, P. E., and Engle, E. T., *Am. J. Anat.,* 1927, **40**, 159.

TABLE I.
Effect of Injections of Pituitary Glands Obtained from Lithosperm and Control Donors on Organs of 21-day-old Mice.

Group	No. mice	Organ weight in mg			Body weight in g	
		Ovaries	Uterus + vagina	Adrenals	21 days old	26 days old
I. Lith. donors	9	5.0 ± 0.3	14.6 ± 1.1	2.4 ± 0.2	6.9 ± 0.5	9.0 ± 0.7
II. Control donors	9	5.8 ± 0.4	26.2 ± 4.0	2.7 ± 0.2	6.9 ± 0.2	9.5 ± 0.3
III. Uninjected controls	10	4.8 ± 0.3	13.5 ± 1.7	1.9 ± 0.2	6.9 ± 0.6	8.6 ± 0.9

not seen in mice receiving pituitary injections from Lithosperm donors (Group I) inasmuch as there is no significant difference between the weights of the uterus plus vagina in these mice and in the uninjected controls (Group III). On the other hand considerable gonadotropic stimulation was evident in the mice receiving pituitary injections from control donors (Group II), the uterus plus vagina weighing 95% more than that of the uninjected controls and 79% more than that of the Lithosperm donor. These differences are significant statistically. Thus the results show that the gonadotropic potency of pituitary glands of adult female mice receiving Lithosperm was decidedly less than that of similar control mice.

No significant differences in the weights of ovaries or adrenal glands or in body weight between the 3 groups were obtained. Had the mice been injected with more pituitary glands, some differences might have been noted, at least in ovarian weight. Increase in the weight of ovaries is also an indicator of gonadotropic stimulation, but a less sensitive one than that of the uterus.[4]

The concentration of the sample of Lithosperm (20%) used in this experiment is a threshold one, producing continuous diestrous in 55 per cent of mice of a sensitive strain (C_3H). A concentration of 30% is required to produce continuous diestrous in all such mice.

Discussion. The finding that Lithosperm decreases the gonadotropic potency of the pituitary gland aids in the understanding of the mechanism of action of this crude drug. The contention that the action is not a direct one on the vagina, ovaries or circulating gonadotropin but involves rather the gonadotropic activity of the pituitary gland is strengthened. Zahl[5] has shown that no histological changes are evident in the pituitary gland after prolonged Lithosperm administration and that normal estrous cycles are reestablished quickly after discontinuance of the drug, even after 8 months of therapy. Thus it appears that Lithosperm inhibits in some way the formation of gonadotropic hormone without producing irreversible or histological damage to the pituitary gland.

Conclusion. The gonadotropic potency of pituitary glands of adult female mice is decreased by the administration of *Lithospermum ruderale* as evidenced by a lack of increase in weight of the uterus plus vagina in 21-day-old mice which have been injected with pituitary glands from treated mice.

[4] Levin, L., and Tyndale, H. H., *Endocrin.*, 1937, **21**, 619.

[5] Zahl, P. A., PROC. SOC. EXP. BIOL. AND MED., 1948, **67**, 405.